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Oversummering of Fire-Blight Pathogen, Spraying for Control of Fire Blight, and Abscission Induced by Erwinia amylovora and Phytomonas syringae

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OVERSUMMERING OF FIRE-BLIGHT PATHOGEN, SPRAYING FOR CONTROL OF FIRE BLIGHT, AND ABSCISSION INDUCED BY ERWINIA AMYLOVORA AND PHYTOMONAS SYRINGAE

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OVERSUMMERING OF THE FIRE-BLIGHT PATHOGEN WITHIN BLIGHTED TISSUES

Numerous studies have been made of the manner in which the fire-blight parasite, *Erwinia amylovora (Bacillus amylovorus)*, passes through the winter *(4, 17, 24, 26, 36)* and how it produces disease during the early part of the growing season. In contrast with these, surprisingly little has been heretofore recorded of studies which trace the life of this destructive parasite through the growing season. Where the growing season is comparatively short, as in the northern United States and in Canada, this may not be an important problem, but where it is 8 or 9 months long, as in Arkansas and throughout the Southern and Pacific states, the problem becomes one of outstanding importance. Notwithstanding this relatively long period of growth of various susceptible pomaceous hosts in Arkansas, the actual period of parasitic activity and production of disease is almost wholly limited to the early part of the season. Rarely are any fresh infections found after the middle of June in this state. The maximum number of infections and greatest degree of invasion on orchard trees occurs usually in April and May. Without doubt this restriction of the disease is largely correlated with the growing condition of the host, as numerous investigators have pointed out. Only during the blossoming period and when the leaves and twigs are rapidly expanding and making noticeable increase in growth, does fire blight of blossoms, twigs, and limbs become serious.[1]

Numerous attempts by the writer to produce disease by injecting virulent cultures of bacteria during the summer or fall into apple and pear twigs of susceptible varieties, which have ceased elongating, have in most instances failed to produce infections when similar attempts made on actively elongating twigs have in almost all instances succeeded.

While the period of maximum disease development corresponds in general with the period of most active vegetative growth, there still remains an interval of 5 to 6 months of growing season

[1]Where collar blight and root blight, associated with Er. amylovora, are present, as in the Pacific and a few eastern states, the period of active expansion of the disease in these tissues may be different from the period of blight development in the upper parts of the trees. This, however, has not been demonstrated.

in Arkansas when very few or no new infections occur, and when the downward or upward extension of blight of affected leaves, twigs, and limbs has ceased either temporarily or entirely. In contrast with certain other bacterial diseases of plants, such as the bacterial spot of peaches and other stone fruits induced by *Phytomonas pruni*, and the angular leaf spot of cotton induced by *P. malvacearum*, in which infections are not limited to the early part of the growing season, the problem of oversummering of *Er. amylovora* is much different and considerably more complex. What happens to the fire-blight pathogen when it is no longer active parasitically, during the relatively hot and frequent dry periods characteristic of the summer months in the Ozarks of Arkansas and Missouri? It is to be noted that summer day-temperatures of 33° to 40° C. (90° to 104° F.) are common in this section, and that according to D. H. Jones *(10)*, Stewart *(32)*, L. R. Jones *(11)*, and E. F. Smith *(31*, p. 372*)*, the optimum temperature for growth is between 22° and 30° C., and the thermal death point of this parasite is between 43.7° to 47° C.[2]

Conceivably there are several ways in which the bacteria may live through the summer. First, and the most important way, as conceived by numerous investigators, they will live within tissues which had become diseased in the early part of the growing season; second, they will live as extruded ooze either on the surface of the diseased tissues or on any other plant parts to which the exudate may have been carried by rain, wind, insects, birds, or other animals, or on and in the soil beneath the affected trees; and third, they will live externally or internally on insects or other animal life which come in contact with the exudate, or in the living quarters, such as beehives, nests, etc., into which the bacteria have been carried.

In order to determine the life span and prevalence of *Er. amylovora* within diseased tissues during the summer and fall seasons, attempts were made to isolate the organism at different times from large numbers of diseased twigs and limbs of pear trees located in three different areas, all in northwest Arkansas. One area involved a group of severely blighted Kieffer pears located close to the laboratory at Fayetteville (see Figure 1);

[2]So far as the thermal relationships on artificial media are concerned, the writer's studies are inconclusive. Three different isolates of **Er. amylovora** were transferred, in comparable quantities of inoculum into tubes of sterile water kept at 43°, 44°, 45°, and 46° C. for 10 minutes. Measured quantities were then transferred into nutrient dextrose broth which was kept at a uniform temperature of 25° C. for several days. The results were inconsistent, growth appearing in some tubes representing transfers from the 45° and 46° treatments, when no growth occurred in any of the tubes representing the 44° treatment. A duplication of the experiment again yielded conflicting data. It is probable that some uncontrolled factor produced these results, and it is also possible that the use of sterile water as the medium for testing the influence of temperature, contrasted with the use of nutrient broth by previous investigators, may explain the difference in results. Sterile water was used by the writer instead of nutrient broth to avoid, in part, the objection of using such a wholly artificial medium as peptone beef-extract broth which has no counterpart in the life of the organism under natural conditions.

Figure 1. Severely infected Kieffer pear trees close to the laboratory at Fayetteville, Arkansas, that were used for part of the studies on the oversummering of the fire-blight bacteria. Owing to the fact that black and white photographs do not clearly distinguish between brown, dead tissue and green, healthy tissue, this one hardly shows the very large amount of disease present on these trees, though the diseased and partly withered leaves seen on the extreme upper twigs against the sky indicate in some measure the amount of disease. Photographed May 9, 1934.

another involved two trees at the Arkansas Experiment Station farm, about 1½ miles away; while the third consisted of a relatively large pear orchard (mostly Kieffers and Garbers) near Bentonville, about 25 miles from the laboratory.

These studies were almost wholly confined to Kieffers and Garbers because no other varieties were grown in this region in sufficient numbers to be of any particular significance with reference to serving as important sources of inoculum. Very rarely were any varieties other than those mentioned found in the intensive fire-blight survey conducted in previous years in the region under consideration *(26)*, and, in these rare instances, the quantity of disease and the types of invasion were quite similar to those observed in Kieffers and Garbers. If Bartlett pears or any other variety were grown which may be more conducive to continued internal invasion by the parasite, it is conceivable that the life span of the bacteria might be different.

The specimens were gathered throughout the summer and fall of 1934 and immediately used for isolation purposes or stored for a few days in a refrigerator, the aim being to isolate the pathogen as soon after the material was gathered as possible. Most investigators have found that *Er. amylovora*, if at all present and viable, can usually be located at the distal end of the blighted portion of the twig or limb, at the margin where healthy and diseased tissues meet *(10, 4, 36, 30)*. Consequently, a portion of the twig or limb, consisting of both living and blighted tissues, was taken in each instance. It was washed in water, momentarily dipped in 95 per cent ethyl alcohol, steeped in 1 to 1,000 mercuric chloride for one to 2 minutes, washed in three changes of sterile water, mashed either in sterile water or nutrient broth, and used for a series of poured dilution plates with nutrient dextrose agar as the medium. This method had previously been proved highly efficient in isolating *Er. amylovora*. After several days' incubation at 28° C., single colonies which in any way resembled *Er. amylovora* were transferred to nutrient broth, permitted to multiply for 3 to 4 days, and then injected with a hypodermic syringe into several succulent healthy shoots of Bartlett pear trees maintained in a greenhouse. If these became infected, the organism was reisolated and single-colony transfers again used for inoculating healthy Bartlett shoots. The following table presents the results of the successful and unsuccessful attempts at isolating the pathogen from the material gathered during the late summer and early fall.

TABLE 1. ISOLATIONS OF *Erwinia amylovora* FROM BLIGHTED PEAR TWIGS AND LIMBS IN THE SUMMER AND FALL OF 1934

Month	Number of twigs and limbs used for isolations								Number of successful isolations				
	Total	Diameter of twigs and limbs in inches							Total	Canker margins		Diameter of tissue in inches	
										Cracked	Uncracked		
		1-8	3-16	1-4	5-16	3-8	1-2	3-4				1-8	3-16
July	254	93	91	52	14	2	2		5	2	3	5	
August	383	79	96	97	48	30	32	1	3	3		3	
September	165	25	45	50	15	14	15	1	None				

In conjunction with the attempt to determine the life span of the organism within blighted twigs and limbs in any one growing season, it is, of course, necessary to consider the various climatic and other factors operative during that particular year which may have an influence on this organism. Figure 2 presents the daily maximum and minimum temperatures and the amount of precipitation in the period under consideration at Fayetteville (University of Arkansas Experiment Station Farm), taken from the Climatological Data, U. S. Department of Agriculture Weather Bureau. The data for Bentonville, the other locality in which the diseased material was gathered, are not given because they closely match those of Fayetteville.

It is to be noted that all sizes of blighted twigs and limbs to be found on the trees were used in these studies, and that a special effort was made to include those of the largest diameter. The fact that no limbs larger than three-fourths of an inch in diameter were used means that no infections of this year's origin on larger limbs were found. In two cases representing an unknown variety at the Arkansas Experiment Station Farm, where the infections had spread from a blighted limb into the main trunk, attempts to isolate the pathogen in July from the margin of the trunk cankers resulted in failure. However, as no effort was made to cut through the entire trunk, the evidence for the presence or absence of viable bacteria in these cases is not considered conclusive. The writer has previously shown *(24)* that the bacteria may at times reside in the xylem of infected tree parts and this has been confirmed by Shaw *(30)*.

Whatever a study of the xylem may have revealed in these cases, it is necessary to direct attention to the fact that continued extension of limb and trunk cankers during the growing season

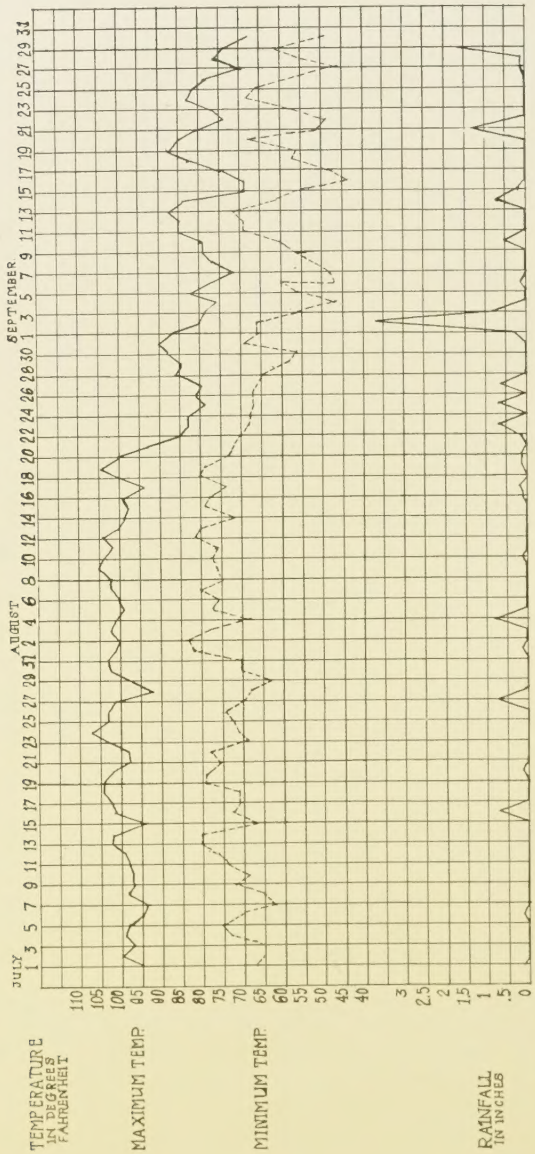

Figure 2. Climatological data for the period involved in the studies of oversummering of the fire-blight bacteria.

and from one year to another, as described on susceptible pears by numerous investigators in northern and Pacific states, is very rare in Arkansas on either pears or apples. Contrasted with reports of bearing trees being killed in one or a few years by extending cankers on trunks and on roots, *(5, 22, 33, 37)*, attacked apple or pear trees of susceptible varieties commonly grown in this state usually linger for many years in spite of repeated attacks of fire blight. The usual appearance here of pear trees that have been grown for 10 years or more is that of large irregular trees with many dead limbs and branches consisting of wood in varying stages of decay and disintegration, or of severely pruned trees with large trunks and relatively short, compact tops. Pear trees 40 or more feet in height and over 20 years of age, and full of blighted and dead limbs are not at all unusual (see Figure 1). Very rarely will a single trunk canker extend completely around a tree and kill it in relatively short order, as occurs in other parts of the country. Indeed, judging by the gross appearance of attacked trees, including blighted and dead limbs in varying stages of decay, it is obvious that individual or seasonal attacks are quite limited in extent of invasion, and that the many dead limbs represent a succession of attacks rather than one or a few which involve extensive trunk or limb invasion.

The data presented show that in no twig or limb studied were the bacteria able to survive in the summer of 1934 in the region considered. Out of 165 isolations made in September, none yielded viable and infectious fire-blight bacteria, contrasted with 5 successful isolations made in July out of 254 blighted twigs and limbs, or slightly less than 2 per cent, and 3 successful isolations out of 383 made in August (0.78 per cent). Clearly, the chances of this parasite living within blighted twigs and limbs through the summer are very limited. These studies indicate more clearly than those previously reported by the writer *(26)* that in this section of the country the survival of the bacteria within blighted tissues from season to season is quite limited when compared with more northern and extreme western fruit-growing regions. While the studies previously reported mainly concerned the common susceptible apple varieties, particularly the Jonathan, and were largely confined to material gathered in the winter, those now presented for the common pear varieties grown in this section add to the knowledge of the longevity of bacteria in blighted wood in this region.

While the data here reported involved a hot and dry period of exceptional duration, which may explain the total failure to obtain viable bacteria in September, there is no good reason for believing that the longevity of bacteria within blighted twigs and limbs is greatly different in the ordinary, or average, Ozark sum-

mer. The ordinary summers are usually hot during the day and often injuriously dry so far as cultivated plants are concerned. With reference to the degree of dryness and its possible lethal effect on this species of bacteria, the writer has shown *(29)* that the bacteria contained in fire-blight exudates can withstand exceedingly dry conditions for at least a year and remain viable and infectious. How long they may live within tissues under measured degrees of dryness remains to be determined. In Arkansas, the relatively short life of *Er. amylovora* within blighted twigs and limbs is probably not due to actual dryness or to high temperatures, though much remains to be determined about the influence of various temperatures on the longevity of this organism.

Whatever the cause of the short life of the bacteria within blighted tissues may be, and there are several possible explanations, it is clear that conditions in Arkansas are such that blighted twigs and limbs are not nearly so important in serving as sources of inoculum for the next season's blight as in more northern regions with shorter growing seasons.

Notwithstanding the complete failure to isolate this organism from any diseased tissues in September, 1934, fire blight appeared in the early part of the 1935 growing season in each one of the three orchards in which these studies of oversummering were made.

INVESTIGATIONS CONCERNING ATTEMPTS TO PREVENT FIRE-BLIGHT INFECTIONS BY MEANS OF SPRAYS

In previous publications *(25, 26)* the writer has presented the results of preliminary studies dealing with attempts to prevent or control fire blight by means of sprays. The studies here reported constitute a report of progress of these investigations and also a report of an attempt to correlate the amount of blight on any one tree or group of trees in any one season with that of individual trees and of a whole orchard in the previous year.

Spray Investigations of 1933

Following the very successful control in 1932 of blossom blight, and to a considerable extent twig blight, by the use of a weak Bordeaux mixture in an isolated Jonathan orchard located near Ozark, Arkansas *(26,* p. 90-93*)*, this orchard was used for similar investigations in 1933. The same block of Jonathans was used as in the previous year, with most of the poorly growing or dead trees cut and removed from the orchard. Thus, there were available 143 Jonathans, all 23 years old but varying a great deal

in size, standing in 11 adjoining rows. Four of these rows, comprising 56 trees, were used as checks and received the customary early season lime sulphur sprays. The remaining seven rows, comprising 87 trees, were used to ascertain the value of weak Bordeaux early season sprays for the control of fire blight.

During the winter the usual pruning of blighted and dead wood was practiced, although the removal of all the former year's blighted twigs was by no means accomplished. However, a very thorough search made on every tree on April 1, 1933, for newly produced bacterial exudate and for renewed activities of old blighted wood that had escaped excision, failed to reveal any signs of such bacterial activities. Despite their age, the trees could be thoroughly examined, because most of them were relatively small and had very open tops. Therefore, it can be stated with a con-

TABLE 2. PERCENTAGE OF PRIMARY BLOSSOM BLIGHT ON CHECK TREES, TUBB'S ORCHARD, NEAR OZARK, ARKANSAS

Tree Number	Row 1	Row 2	Row 3	Row 4
	Per cent	*Per cent*	*Per cent*	*Per cent*
1	87	94	60	9
2	65	62	31	40
3	84	91	22	7
4	62	99	10	10
5	50	96	10	4
6	51	79	0*	10
7	31	34	4*	20
8	20	27	2*	29
9	30	29	0*	20
10	21	17	9*	16
11	12	2	1*	33
12	15	13	6*	
13	4	8	8	
14		1	45	
15		5		
16		1		
17		3		
18		18		
Mean Percentage	40.9	40.3	14.8	18.0

*Tree dying.

siderable degree of assurance that renewed activity of any overwintered blight was not overlooked. The number of overwintered blighted twigs remaining on the trees was quite limited, and they were easily detected on April 1. On that date floral and leaf formation on all living wood had advanced to the point where the flower buds were commencing to show separation in the clusters, and many of the leaf buds were partly expanded and green.

The fire blight which subsequently appeared in this orchard bore no relationship to overwintered blight on any one tree or group of trees, and was fairly uniformly scattered throughout the whole check plot of 56 trees, as may be seen in Table 2. As soon as the first blight made its appearance, a very careful effort was made to trace it to nearby blighted material of the previous season. Any previously diseased twig or limb found in the vicinity of the new blight was not only examined in the orchard by cutting into the healthy wood at the margin of last year's blight and looking for signs of fresh extensions, but was brought into the laboratory and attempts were made to isolate the parasite. In no instance was evidence obtained of the presence of living fire-blight bacteria in the old blighted material.

The weak Bordeaux spray, consisting of one pound of powdered copper sulphate and 3 pounds of hydrated lime to 50 gallons of water, was applied on the following dates:

April 5. Cluster bud spray.
April 11. First open-blossom spray when approximately 25 per cent of the blooms were wide open.
April 19. Second open-blossom spray when approximately 75 to 80 per cent of the blooms were wide open.
May 2. Calyx spray.
May 19. First cover spray.

The check trees, receiving $1\frac{1}{2}$ gallons of commercial lime sulphur to 50 gallons of water, were sprayed on the same dates as those receiving the Bordeaux mixture, with the exception that the checks received no open-blossom sprays. Arsenate of lead, at the rate of $1\frac{1}{2}$ pounds to 50 gallons of spray, was added to the calyx spray and the subsequent cover sprays.

The first fire blight, in the form of blossom infection, was found on April 20 on one of the check trees. It consisted of a reddish-brown discoloration of a number of filaments borne in one blossom, the discoloration being most conspicuous toward the base. As microscopic examination of one of the discolored filaments revealed great numbers of bacteria, a number of fila-

Figure 8. Infected Bartlett pear shoots typical of all shoots artificially inoculated with single-colony transfers representing isolations from diseased filaments of Jonathan stamens. Photographed 3 days after the inoculations were made.

ments similarly discolored and borne on the same blossom were used for isolation purposes in a series of poured dilution plates. From characteristic colonies which appeared in great numbers on the plates, single-colony transfers were made and used for inoculation on healthy succulent Bartlett pear shoots on trees maintained in a greenhouse (Figure 3). All inoculated shoots developed typical fire-blight symptoms within a few days after inoculation. All uninoculated shoots remained healthy.

No other blighted blossoms or any other form of blight was noted on April 20, but on April 29 a number of blossom clusters were noted with initial signs of blight and a few leafy shoots were found blighted. The amount of disease was rather difficult to count accurately at this time, because the symptoms were, as a whole, only slightly visible.

By May 8 primary blight had developed to its fullest extent and could easily be counted (see Table 2). The secondary blight, mostly in the form of internal extensions from the primary infections, was not in evidence until a later date. However, the secondary blight was so insignificant in amount that it is not recorded.

The percentage of blossom blight, as seen in Table 2, varied a great deal from tree to tree, though the variation was probably no greater than that involved in the marked difference in the total number of blossom clusters borne on the different trees. There was fair indication of a direct correlation between the total number of blossom clusters borne on any one tree and the number blighted. In general, those trees which had few clusters had a relatively small percentage of blight, while those with many had proportionately larger percentages of the disease. Likewise, in a number of instances, as in Row 3, Trees No. 6 to 13, where a small percentage of blossom blight occurred, all of the trees were in the process of dying. The death of such trees was apparently due to improper soil conditions plus an extreme amount of sunscald on the trunks and larger limbs.

The observations suggested that the difference in per cent of blight was not so much due to a difference in available inoculum or to a difference in degree or kind of dissemination, but to a difference in the degree of susceptibility of different trees. As a whole, those trees which showed extreme stunting and were otherwise debilitated possessed a relatively small per cent of blight, while those of greater vigor and greater fruitfulness possessed more blight. There was, however, a number of trees in which no correlation was noted between vigor or amount of blossom production and per cent of disease. This, of course, does not exclude the possibility of a connection in such cases be-

tween some variable physiological or morphological host character and the amount of disease inception.

In contrast with the relatively large amount of blossom blight on the check trees as a whole, the 87 Bordeaux-sprayed trees had less than one per cent of the blossom clusters blighted. The total number of blighted clusters in the whole treated block was 92, an insignificant number when compared even with individual trees among the checks; for example, Tree No. 4, Row 2, had 2,805 blighted blossom clusters.

From a practical viewpoint, the control of fire blight in this orchard in 1933 by the use of weak Bordeaux sprays was nearly perfect, and closely duplicated the results obtained in the same orchard in 1932. Not only was fire blight controlled almost perfectly, but the Bordeaux-sprayed trees, as a whole, looked thriftier, the leaves appeared deeper green in color, and owing to the control of the disease, were present in far greater numbers than on the check trees (compare Figure 4 with Figure 5).

While it was not possible to make accurate counts of the influence of the two full-blossom sprays on the setting of fruit, there was no indication that the Bordeaux sprays had adversely influenced the setting. On the contrary, there was a marked suggestion of a better setting of fruit on the Bordeaux-sprayed trees than on those sprayed with lime sulphur, irrespective of the amount of blossom blight. As to the amount of spray injury on the fruit, those sprayed with Bordeaux mixture showed russeting of slightly less than 9 per cent of the apples, and most of this appeared on the fruit borne on the north side of the trees. The fruit sprayed with lime sulphur showed little russeting or scalding, and was, as a whole, more highly colored. The amount of russeting present on the Bordeaux-sprayed fruit was of no consequence from an economic viewpoint as the particular markets served paid no premium for the smoother or more highly-colored fruit.

Spray Investigations of 1934

To obtain results with protective sprays which are capable of being duplicated, it is obvious that some knowledge must be had of the amount of inoculum and of other factors conducive to infection operative in any orchard used for spray investigations.

For this reason the apple orchard used for spray investigations in 1934 and in 1935 was thoroughly studied for cases of possible overwintering cankers, and for signs of activities of the

Figure 4. Check (lime sulphur-sprayed) tree with many blossom clusters blighted, characteristic of the checks as a whole. Photographed at Tubb's orchard, Ozark, Arkansas, May 22, 1933. (Compare with Figure 5.)

Figure 5. Bordeaux-sprayed tree, characteristic of 87 trees sprayed similarly showing the absence of blossom and twig blight. Photographed at Tubb's orchard, Ozark, Arkansas, May 22, 1933. (Compare with Figure 4.)

fire-blight bacteria. The particular portion of this orchard[3] used for spray investigations consisted of a solid 6-row block of Jonathans of 167 trees, which were 12 years old in 1934. This block was severely blighted in 1933, the year previous to the initiation of the studies about to be reported. In 1933 the disease was so serious on many of the trees, particularly those on the east side of the orchard, that fully 75 per cent of the crop was lost.

It may be desirable to call attention to the type of soil and general arrangement of this orchard. The soil is one generally classified as Clarksville silt loam, consisting of about 8 to 12 inches of a grayish silty top soil over a brownish clay subsoil at the north end, and somewhat shallower top soil with more of a bluish-brown clay subsoil at the south end with obviously poorer drainage at this end. The subsoil at both ends and toward the middle possesses a relatively large quantity of chert. The block of Jonathans utilized for the tests was bordered on the east by an open field utilized for garden, meadow, and various cultivated annuals; on the south by a non-orchard area used for various plants; on the west by several rows of Staymans followed by additional apple rows of Red Delicious, and on the north by a road bordering another apple orchard. This north orchard, as well as the rows of Staymans and Red Delicious within the orchard utilized for spray investigations, had no recognizable fire blight up to the present (1936). The blight-susceptible Jonathan block had shown great variation in the amount of fire blight. Invariably, the extreme west row (Row 6), bordering the Staymans, had for some unaccountable reason suffered very little blight compared with the remaining 5 rows. This was true in 1933, the year previous to the utilization of this orchard for spray investigations, as well as in the two succeeding years.

Attempts to correlate the amount of disease present in one season with that of the previous season. In order to determine the relative amount of inoculum which might be produced by the blighted and overwintered materials left on the trees, a very thorough count of all blighted blossom clusters, twigs, and limbs was made on each tree during the late winter and early spring of 1934 after the trees had received a light pruning. In spite of this pruning, the amount of blighted material left on the trees was so great, as shown in Table 3 and Figure 6, that if any correlation exists between the number of overwintered blighted

[3]This orchard, owned at the time by J. W. Leazer, is located about 2 miles northwest of Rogers, Benton County, in an area extensively used for apple growing.

TABLE 3. COUNTS OF OVERWINTERED BLIGHTED BLOSSOM CLUSTERS, TWIGS, AND LIMBS MADE IN FEBRUARY, MARCH, AND EARLY APRIL, 1934, IN LEAZER'S ORCHARD, NEAR ROGERS, ARKANSAS

Row number	Tree number	Blighted blossom clusters	Blighted twigs ½ inch or less in diameter	Blighted twigs ½ inch to 1 inch in diam.	Blighted limbs 1 inch or more in diam.	Total blighted twigs and limbs per tree
		Number	*Number*	*Number*	*Number*	*Number*
1	1	154	210	25	10	245
1	2	120	221	25	10	256
1	3	92	216	27	9	252
1	4*	2	21	0	0	21
1	5	186	472	12	8	492
1	6	282	740	43	9	792
1	7	293	279	20	3	302
1	8	54	74	1	3	78
1	9	161	177	8	4	189
1	10	108	83	4	3	90
1	11	95	227	2	3	232
1	12	0	9	0	0	9
1	13	60	169	3	4	176
1	14	48	54	1	0	55
1	15	54	191	3	0	194
1	16	3	39	2	1	42
1	17	31	83	6	0	89
1	18	18	50	3	0	53
1	19	32	49	4	0	53
1	20	vacant				
1	21	55	130	2	1	133
1	22	74	110	4	1	115
1	23	35	84	9	1	85
1	24	3	4	0	0	4
1	25	9	7	0	0	7
1	26	13	11	0	0	11
1	27	16	19	0	0	19
1	28	15	38	2	0	40
1	29	41	77	2	0	79
1	30	vacant				
1	31	vacant				
Totals		2054	3844	199	70	4113
2	1	37	95	0	0	95
2	2	132	205	2	0	207
2	3	98	183	5	0	188
2	4	133	129	4	0	[illegible]
2	5	262	329	7	1	[illegible]
2	6	126	229	1	1	[illegible]
2	7	vacant				
2	8	84	91	3	1	95
2	9	140	103	0	1	104
2	10	54	84	6	1	91
2	11	63	88	1	1	90
2	12	42	87	2	0	89
2	13	37	66	1	0	67
2	14	10	11	1	0	12
2	15	50	72	1	0	73
2	16	37	41	1	0	42
2	17	12	13	1	0	14
2	18	8	11	0	0	11
2	19	40	35	0	0	35
2	20	53	43	0	0	43
2	21	17	24	0	0	24
2	22	33	18	0	0	18
2	23	11	30	0	0	30
2	24	12	26	3	0	29
2	25	19	27	1	0	28
2	26	8	15	0	0	15
2	27	16	52	2	2	[illegible]
2	28	33	40	0	0	40
2	29	27	28	1	0	29
2	30	12	20	0	0	20
2	31	0	4	0	0	4
Totals		1606	2179	43	8	2230

*Not Jonathan.

TABLE 3. COUNTS OF OVERWINTERED BLIGHTED BLOSSOM CLUSTERS, TWIGS, AND LIMBS MADE IN FEBRUARY, MARCH, AND EARLY APRIL, 1934, IN LEAZER'S ORCHARD, NEAR ROGERS, ARKANSAS—(*Continued*)

Row number	Tree number	Blighted blossom clusters	Blighted twigs ½ inch or less in diameter	Blighted twigs ½ inch to 1 inch in diam.	Blighted limbs 1 inch or more in diam.	Total blighted twigs and limbs per tree
		Number	*Number*	*Number*	*Number*	*Number*
3	1	43	42	0	0	42
3	2	98	83	5	2	90
3	3	139	91	2	0	93
3	4	126	135	4	0	139
3	5	377	143	2	1	146
3	6	198	125	4	0	129
3	7	58	42	0	0	42
3	8	139	187	3	0	190
3	9	vacant				
3	10	27	86	4	1	91
3	11	41	64	1	0	65
3	12	21	42	1	0	43
3	13	28	52	0	0	52
3	14	88	118	0	0	118
3	15	64	83	0	0	83
3	16	6	2	0	0	2
3	17	24	16	0	0	16
3	18	9	20	0	0	20
3	19	40	18	0	0	18
3	20	6	1	1	0	2
3	21	9	33	0	0	33
3	22	11	3	0	0	3
3	23	7	6	0	0	6
3	24	19	12	1	0	13
3	25	42	40	1	0	41
3	26	57	88	1	2	91
3	27	vacant				
3	28	vacant				
3	29	8	6	0	0	6
3	30	4	2	1	1	4
3	31	15	26	0	2	28
Totals		1702	1566	31	9	1606
4	1	96	69	5	1	75
4	2	vacant				
4	3	106	110	2	0	112
4	4	146	193	4	0	197
4	5	66	43	0	0	43
4	6	6	5	0	0	5
4	7	189	170	2	0	172
4	8	dead				
4	9	89	24	0	0	24
4	10	20	20	0	0	20
4	11	66	76	1	0	77
4	12	44	58	0	0	58
4	13	11	22	1	0	23
4	14	25	33	1	1	35
4	15	14	14	0	0	14
4	16	36	42	2	0	44
4	17	15	30	1	0	31
4	18	30	45	4	1	50
4	19	37	79	2	0	81
4	20	dwarfed				
4	21	16	13	0	0	13
4	22	14	19	0	0	19
4	23	22	20	1	0	21
4	24	17	10	0	0	10
4	25	11	10	0	0	10
4	26	35	35	1	0	36
4	27	20	12	0	0	12
4	28	vacant				
4	29	vacant				
4	30	9	21	0	0	21
4	31	5	11	0	1	12
Totals		1145	1184	27	4	1215

TABLE 3. COUNTS OF OVERWINTERED BLIGHTED BLOSSOM CLUSTERS, TWIGS, AND LIMBS MADE IN FEBRUARY, MARCH, AND EARLY APRIL, 1934, IN LEAZER'S ORCHARD, NEAR ROGERS, ARKANSAS—(*Continued*)

Row number	Tree number	Blighted blossom clusters	Blighted twigs ½ inch or less in diameter	Blighted twigs ½ inch to 1 inch in diam.	Blighted limbs 1 inch or more in diam.	Total blighted twigs and limbs per tree
		Number	*Number*	*Number*	*Number*	*Number*
5	1	118	191	6	0	197
5	2	68	69	0	0	69
5	3	67	74	3	0	77
5	4	63	49	1	0	50
5	5	123	125	1	0	126
5	6	85	46	0	0	46
5	7	269	218	1	0	219
5	8	[illegible]	66	2	1	68
5	9	84	38	0	0	38
5	10	92	72	5	1	78
5	11	32	17	0	0	17
5	12	33	19	0	0	19
5	13	34	32	1	0	33
5	14	15	28	0	1	29
5	15	17	14	1	1	16
5	16	33	7	0	0	7
5	17	67	42	0	0	42
5	18	14	16	3	0	19
5	19	18	25	0	0	25
5	20	18	15	0	0	15
5	21	15	4	0	0	4
5	22	17	15	2	0	17
5	23	9	21	0	0	21
5	24	16	20	1	0	21
5	25	23	20	1	0	21
5	26	9	9	1	0	10
5	27	vacant				
5	28	vacant				
5	29	dead				
5	30	5	1	0	2	3
5	31	5	5	0	0	5
Totals		1404	1258	29	6	1292
6	1	48	43	0	0	43
6	2	38	57	2	1	60
6	3	40	41	0	1	42
6	4	41	69	3	0	72
6	5*	3	4	0	0	4
6	6	21	19	0	0	19
6	7	81	67	1	0	68
6	8	21	16	0	0	16
6	9	27	37	2	0	39
6	10	8	6	0	0	6
6	11	20	30	0	0	30
6	12	25	43	0	0	43
6	13	6	8	0	0	8
6	14	7	10	0	0	10
6	15	16	16	0	0	16
6	16	53	37	1	0	38
6	17	28	53	1	1	55
6	18	2	6	0	0	6
6	19	14	13	0	0	13
6	20	16	14	0	0	14
6	21	15	7	0	0	7
6	22	dwarfed				
6	23	27	34	0	0	34
6	24	0	3	0	0	3
6	25	5	2	0	0	2
6	26	0	1	0	0	1
6	27	vacant				
6	28	vacant				
6	29	0	0	0	0	0
6	30	14	23	0	0	23
6	31	11	12	0	0	12
Totals		[illegible]	[illegible]	[illegible]	3	684

*Not Jonathan

Figure 6. Jonathan apple trees, Leazer's orchard near Rogers, Arkansas, used for spray investigations for the control of blossom blight. Note the many leafless twigs representing, in most instances, wood that had been blighted in the previous year. Photographed April 21, 1934.

twigs and limbs, and the severity of blight in the succeeding year, there was ample opportunity of observing and recording this relationship.

Table 3 shows not only the large amount of overwintered blighted material of various types in the Jonathan block as a whole, but indicates, as previously noted, that the trees in Row 6 were much less subject to blight in 1933 than any of the other rows of trees. The same thing will be noted in subsequent years.

For the spray investigations looking toward the prevention

of fire blight, and particularly the blossom blight stage of this disease, the block was divided as follows:

Row 1. Copper phosphate mixture with two open-blossom sprays.
Row 2 (check). Copper phosphate mixture without open-blossom sprays.
Row 3 (check). Bordeaux mixture 1-3-50 without open-blossom sprays.
Row 4 Bordeaux mixture 1-3-50 with two open-blossom sprays.
Row 5 (check). Lime sulphur with two open-blossom sprays.
Row 6 (check). Lime sulphur without two open-blossom sprays.

Based upon the results obtained previously in the control of this disease *(25, 26)* the spray plot was so arranged that Rows 1 and 4, those receiving two open-blossom copper sprays, in addition to the ordinary cluster-bud and calyx sprays used in this section in the early part of the season, were expected to show reduced amounts of blossom blight, while Rows 2, 3, 5, and 6 were to serve as controls. Row 5, while receiving two additional sprays of lime sulphur, to compare with the two additional open-blossom sprays of Rows 1 and 4, was not expected to show any appreciable control.

The respective sprays were applied on the following dates:

April 16. Cluster-bud spray.
April 21. First open-blossom spray; approximately 25 per cent of blossoms wide open.
April 25. Second open-blossom spray; approximately 75 to 80 per cent of blossoms wide open.
April 28. Calyx spray.
May 16. First cover spray.

The remainder of the spray schedule simply followed the Station entomologists' recommendations for the control of codling moth, with copper phosphate mixture added in Rows 1 and 2 and Bordeaux mixture 3-4-50 in Rows 3, 4, 5, and 6 for controlling blotch and bitter rot. Since these are not concerned with fire-blight control, they will not be considered further.

The first infections, noted on May 10, were mostly in the form of blossom blight (Figure 7), and from that date until no further signs of additional blight appeared, records were kept of the amount and kind of blight on each tree. It is of interest to note that no new infections were found after the last week in May. Owing to the relatively small amount of fire blight in the block as a whole, there is no particular value in recording the amount

Figure 7. The first form of fire blight found in Leazer's orchard, near Rogers, Arkansas, observed and photographed May 10, 1934. Note the partly withered leaves and the completely killed blossom cluster. On this date the blossom cluster immediately above the one seen, had abscissed, while a third one only partly visible, on the upper right, had also blighted.

and kind of blight noted on the different dates. Likewise, the insufficiency of disease on the check rows does not permit drawing conclusions concerning the value of the open-blossom sprays. It is, however, of importance to note the total counts of blighted blossom clusters and twigs and limbs on each tree, as shown in Table 4. While the data are of little value in determining the value of the sprays for the prevention of fire blight in that season, a comparison of the amount of blight on each tree with the amount of blight on the same tree in the previous season is of considerable interest. Excluding Rows 1 and 4, in which the two additional open-blossom copper sprays may have reduced the number of blossom infections, or including these two rows, there is no correlation to be noted on individual trees or on a group of adjoining trees in any part of the block. Whether one chooses those trees which had the greatest number of blighted twigs and limbs in 1933 and compares them with the amount of blight on the same trees in 1934, or chooses those which had the least amount of blight in 1933 and compares them with the 1934 record of these trees, the same lack of correlation between the amount of blight during the 2 years is to be noted. Thus, trees No. 5, 6, and 7 on Row 1, which in 1933 had 186, 282, and 293 blighted blossom clusters, and 492, 792, and 302 blighted twigs and limbs, respectively, showed the following amounts of blight in 1934: 5, 33, and 21 blighted blossom clusters, and 1, 3, and 1 blighted twigs and limbs. Likewise, trees No. 4, 5, and 6 on Row 2 had 133, 262, and 126 blighted blossom clusters, and 133, 337, and 231 blighted twigs and limbs, respectively, in 1933, and 55, 28, and 24 blighted blossom clusters and 1, 4, and 2 blighted twigs and limbs in 1934. In other words, trees with much blight in 1933 had comparatively little blight in 1934. Conversely, tree No. 19, Row 4, and trees No. 9 and 19, Row 5, which had considerable blight in 1934 compared with other trees in the same year, showed no such preponderance of blight in 1933.

These data are directly in line with previous studies (*26*, p. 14-19) in which no general connection was noted between the prevalence of blight in a given orchard in one season when compared with the succeeding one. It is, nevertheless, totally different from the findings in some of the Northern and Pacific states. Thus, in Wisconsin, Miller (*17*) found a rather striking correlation between the number of holdover cankers and blighted twigs of both Transcendent crab-apple and of Fameuse apple, and the amount of blossom infection; Tullis (*36*), in Michigan, found similar relationships in both apples and pears; in New York, Thomas and Parker (*35*) have found essentially the same relationship; in California, Thomas and Ark (*34*) report similar observa-

TABLE 4. COUNTS OF FIRE BLIGHT ON BLOSSOM CLUSTERS, TWIGS, AND LIMBS, JUNE, 1934, IN LEAZER'S ORCHARD, NEAR ROGERS, ARKANSAS

Row number	Tree number	Blighted blossom clusters	Total cluster set	Blighted twigs and limbs
		Number	*Number*	*Number*
1	1	42	301	5
1	2	29	112	2
1	3	10	111	1
1	4*	0	0	0
1	5	5	87	1
1	6	33	68	3
1	7	21	200	1
1	8	9	15	0
1	9	18	201	0
1	10	19	51	2
1	11	5	287	0
1	12	33	64	5
1	13	12	157	1
1	14	7	209	1
1	15	18	103	1
1	16	7	0	1
1	17	8	261	0
1	18	9	204	0
1	19	11	205	0
1	20	vacant		
1	21	24	444	3
1	22	32	354	1
1	23	5	315	0
1	24	0	23	0
1	25	0	27	0
1	26	0	3	0
1	27	12	84	0
1	28	40	184	5
1	29	2	233	1
1	30	vacant		
1	31	vacant		
Totals		411	4703	34
2	1	8	135	0
2	2	4	87	0
2	3	7	292	0
2	4	55	312	1
2	5	28	122	4
2	6	24	154	2
2	7	vacant		
2	8	35	363	3
2	9	20	159	1
2	10	9	177	1
2	11	6	77	0
2	12	42	218	5
2	13	7	89	5
2	14	0	27	0
2	15	37	229	3
2	16	5	64	1
2	17	1	55	0
2	18	0	3	0
2	19	10	287	0
2	20	28	338	0
2	21	2	196	0
2	22	3	103	0
2	23	87	357	9
2	24	11	180	1
2	25	2	267	0
2	26	0	28	0
2	27	7	223	0
2	28	8	440	3
2	29	5	335	2
2	30	0	80	0
2	31	0	0	0
Totals		451	5397	41

*Not Jonathan.

TABLE 4. COUNTS OF FIRE BLIGHT ON BLOSSOM CLUSTERS, TWIGS, AND LIMBS, JUNE, 1934, IN LEAZER'S ORCHARD, NEAR ROGERS, ARKANSAS—(*Continued*)

Row number	Tree number	Blighted blossom clusters	Total cluster set	Blighted twigs and limbs
		Number	*Number*	*Number*
3	1	9	117	3
3	2	16	154	0
3	3	20	268	5
3	4	8	20	0
3	5	2	20	0
3	6	50	148	3
3	7	2	149	0
3	8	138	181	7
3	9	vacant		
3	10	19	202	1
3	11	2	25	0
3	12	2	27	0
3	13	14	268	0
3	14	31	226	0
3	15	12	399	0
3	16	2	176	0
3	17	6	317	0
3	18	3	177	0
3	19	19	480	1
3	20	2	45	0
3	21	34	389	4
3	22	5	133	1
3	23	1	25	0
3	24	11	179	0
3	25	15	435	2
3	26	32	335	1
3	27	vacant		
3	28	vacant		
3	29	4	232	0
3	30	6	124	0
3	31	5	248	0
Totals		470	5499	28
4	1	11	341	5
4	2	vacant		
4	3	11	156	3
4	4	41	265	5
4	5	2	62	1
4	6	0	99	0
4	7	34	220	5
4	8	vacant		
4	9	21	123	1
4	10	0	11	0
4	11	40	79	9
4	12	11	219	2
4	13	20	444	1
4	14	14	133	0
4	15	0	2	0
4	16	9	7	2
4	17	18	242	2
4	18	35	339	0
4	19	397	232	15
4	20	20	19	1
4	21	7	192	0
4	22	9	262	4
4	23	22	157	2
4	24	5	209	0
4	25	4	81	0
4	26	26	583	2
4	27	4	300	0
4	28	vacant		
4	29	vacant		
4	30	9	173	2
4	31	11	204	1
Totals		781	5154	63

TABLE 4. COUNTS OF FIRE BLIGHT ON BLOSSOM CLUSTERS, TWIGS, AND LIMBS, JUNE, 1934, IN LEAZER'S ORCHARD, NEAR ROGERS, ARKANSAS—(*Continued*)

Row number	Tree number	Blighted blossom clusters	Total cluster set	Blighted twigs and limbs
		Number	*Number*	*Number*
5	1	4	72	2
5	2	5	78	2
5	3	7	99	2
5	4	33	108	10
5	5	37	135	9
5	6	6	29	1
5	7	15	87	3
5	8	79	61	13
5	9	504	116	25
5	10	52	78	4
5	11	22	59	14
5	12	65	135	18
5	13	99	200	11
5	14	1	23	1
5	15	49	118	7
5	16	6	81	0
5	17	35	298	5
5	18	2	1	1
5	19	190	193	3
5	20	17	84	1
5	21	6	16	1
5	22	19	106	3
5	23	56	132	11
5	24	8	28	2
5	25	8	221	0
5	26	dead		
5	27	vacant		
5	28	vacant		
5	29	dead		
5	30	5	111	0
5	31	11	15	2
Totals		1341	2684	152
6	1	10	267	0
6	2	8	129	1
6	3	3	68	0
6	4	28	340	1
6	5*			
6	6	1	28	0
6	7	114	316	0
6	8	7	26	0
6	9	101	353	12
6	10	5	27	1
6	11	57	152	7
6	12	45	263	11
6	13	25	174	6
6	14	13	119	2
6	15	3	103	1
6	16	14	105	1
6	17	48	324	2
6	18	29	257	2
6	19	9	177	2
6	20	15	92	2
6	21	13	118	1
6	22	dwarfed		
6	23	42	561	1
6	24	1	72	0
6	25	4	117	0
6	26	0	0	0
6	27	vacant		
6	28	vacant		
6	29	0	0	0
6	30	dead		
6	31	7	145	1
Totals		602	4253	68

*Not Jonathan.

tions for pears. In other words, given the varieties commonly grown in this region, the possibility of overwintered, blighted twigs and limbs, serving as sources of infection, is not nearly as great in Arkansas and probably in other Southern states, as it is in certain varieties commonly grown in the northern and far western states, despite the fact that fire blight is often far more common and destructive in the South than in the North.

From a practical point of view, the studies here reported help to explain why excision of blighted twigs and limbs has not by itself resulted in satisfactory control of this malady in the Ozarks of Arkansas and Missouri. As indicated in a previous publication (*26*), there are several other possible sources of primary inoculum.

Spray Investigations of 1935

In view of the relatively small amount of fire blight experienced in 1934 in the commercial orchard used for spray investigations, and the necessity of additional trials, the same orchard was again utilized in 1935 for similar studies. The arrangement of the different sprays in relationship to the different rows was exactly similar to that given in 1934, except that Row 6, because of its erratic behavior in former years, as well as in 1935, was excluded from further consideration.[4]

Since the amount of fire blight present in 1934 was relatively insignificant, the amount of pruning necessary to free the orchard of blighted wood was in turn very slight, and while some twigs blighted in 1934 were overlooked in the pruning operation, there were very few left.

The sprays were applied on the following dates:

April 9. Cluster-bud spray.

April 15. First open-blossom spray. While many of the trees had 20 to 25 per cent of the blossoms wide open on this date, a few trees showed as much as 50 to 75 per cent of the blossoms fully open. There was great variation in this respect from tree to tree.

April 18. Second open-blossom spray. Owing to continued cool weather, the blossoms continued a very uneven development

[4]This row, for 3 years in succession, had shown such a little amount of disease that it appeared wholly foreign to the remainder of the Jonathan block. It has already been noted that this row adjoined a disease-free block of Staymans followed by Red Delicious. According to Gossard and Watlow's idea (*9*) that rapidity of pollination has a marked influence on disease initiation in apple blossoms, though this is disclaimed by Pierstorff (*20*), is it possible that the nearness of other varieties, by virtue of a more accessible cross-pollination, is responsible for the relative freedom from disease of Row 6? There is certainly no conclusive evidence for this hypothesis, but if it were true, it offers a notable suggestion for disease prevention, namely, interplanting Jonathans or other susceptible varieties between non-susceptible ones possessing pollen that are compatable with Jonathan pollination.

Figure 8. Infected Jonathan blossoms. The two upper ones and the lower one resulted from pure-culture spray inoculations in the field on April 17, 1934. Photographed 10 days later when the first sure signs of infection had appeared. Diseased blossoms in the photograph appear withered and darker colored than the healthy.

on the different trees. Counts of open blossoms made throughout the orchard showed a variation of 40 to 95 per cent, the greatest number of trees having approximately 75 per cent of the blossoms wide open on this date.

April 27. Calyx spray.

May 7. First cover spray.

As in 1934, the remainder of the spray schedule primarily followed the entomologists' recommendations for the control of codling moth, the arsenate of lead being combined with the copper phosphate spray on Rows 1 and 2, and with 3-4-50 Bordeaux mixture on all the other rows.

Throughout the early part of the growing season, including the blossoming period, a very careful inspection was made of the trees for signs of activity of overwintered blight and especially for signs of bacterial exudate. None were observed. This, of course, was to be expected, since the studies of the oversummering of the parasite in blighted pear wood of 1934, as recorded previously, revealed no case of viable bacteria being present in September in 165 attempts at isolation involving that number of blighted twigs and limbs. Also, various investigators have claimed that pear trees are more likely to serve as sources of overwintering than apples *(24)*.

Notwithstanding the absence of inoculum from blighted wood of the previous season, fire blight appeared relatively early in the apple orchard used for the spray investigations, and as the season advanced, the amount of diseased blossom clusters and twigs, as seen in Table 5, reveals a considerable amount of disease. The earliest signs of blight noted were on April 30, and were confined entirely to the blossoms. No symptoms of disease on leaves or twigs were noted until May 7.

Previous studies *(26)* having shown that an incubation period of 7 to 10 days is usually required under field conditions for clear symptoms to be observable on apple blossoms (Figure 8), it would seem that the infections noted on April 30 occurred in the period between April 20 and 23. The second open-blossom spray, having been applied on April 18, was, therefore, presumably timely enough to protect all blossoms sprayed with germicidal materials that were open on that date.[3] That a comparatively large number of blossoms, varying from tree to tree, had not yet opened by April 18, and, therefore, were unprotected in the vulnerable period of April 20 to 23, probably accounts for some of the variation in percentage of initial blossom blight in the rows receiving the protective sprays. In other words, considering the

[3]It will be shown elsewhere that a common means of entrance of Er. amylovora into apple and pear blossoms is through the exposed stigmatic surfaces, exposed stamens, and nectarial discs of open blossoms.

TABLE 5. TOTAL COUNTS OF BLOSSOM, TWIG, AND LIMB BLIGHT PRESENT IN LEAZER'S ORCHARD, NEAR ROGERS, ARKANSAS, JUNE, 1935

Tree number	Row 5		Row 4		Row 3		Row 2		Row 1	
	Blossom blight	Twig and limb blight	Blossom blight	Twig and limb blight	Blossom blight	Twig and limb blight	Blossom blight	Twig and limb blight	Blossom blight	Twig and limb blight
	Number	*Number*	*Number*	*Number*	*Number*	*Number*	*Number*	*Number*	*Number*	*Number*
1	222	9	54	4	78	4	60	4	220	6
2	123	5	—*	—	29	1	362	8	422	7
3	116	12	97	10	106	3	124	3	446	7
4	55	9	186	7	46	3	208	6	—	—
5	324	18	85	2	62	8	133	4	215	5
6	6	2	—	—	131	9	94	1	199	14
7	483	10	141	5	32	2	—	—	158	1
8	253	9	—	—	424	6	195	5	24	3
9	150	7	14	5	—	—	231	13	23	5
10	55	7	0	0	255	11	176	7	119	1
11	15	3	66	7	26	2	48	2	110	12
12	15	1	91	10	77	5	90	5	17	4
13	162	2	59	4	60	6	154	5	182	2
14	178	7	77	11	325	9	19	0	—	—
15	238	15	11	3	11	3	244	9	63	5
16	60	2	65	5	12	1	151	5	7	6
17	197	3	13	1	103	6	115	13	94	0
18	35	5	3	1	51	5	43	2	60	5
19	145	2	225	9	—	—	163	4	41	5
[illegible]	[illegible]	5	11	0	3		—	—	—	—
[illegible]	[illegible]	6	84	5	144	3	138	6	52	4
[illegible]	[illegible]	4	41	8	36	4	135	8	34	2
[illegible]	[illegible]	7	115	7	18	6	67	3	99	11
[illegible]	[illegible]	12	49	4	31	11	42	4	—	—
[illegible]	80	5	28	2	109	10	85	10	36	3
[illegible]	—	—	145	2	118	18	115	12	0	0
[illegible]	—	—	9	0	—	—	372	7	21	0
[illegible]	—	—	—	—	—	—	185	9	121	11
[illegible]	—	—	—	—	44	5	11	7	165	16
[illegible]	4	1	54	3	60	10	40	8	—	—
51	0	0	57	5	175	12	—	—	—	—
Total	3478	168	1779	120	2566	163	3800	170	2928	135

[illegible] ble to fire blight.

lengthy period involved between the opening of the first and the last blossoms, a period extending from April 9 to April 25, it is clear that it would have taken more than two open-blossom sprays to have fully protected all or nearly all the blooms shortly after they had opened.

Comparing the amounts of blossom blight on the different rows, it will be seen that Row 4, representing all Bordeaux sprays including two open-blossom sprays, has nearly 50 per cent less blight than Row 5 which represents the check lime sulphur treatment, including two open-blossom sprays. Likewise, it appears that the two additional open blossom sprays applied on Row 4 exercised considerable control over Row 3, which also had Bordeaux sprays, but lacked the two open-blossom sprays. A comparison between Row 1, which received copper phosphate sprays, including two open-blossom sprays, and Row 4 seemingly shows considerable difference in favor of the Bordeaux sprays; but when subjected to statistical analysis (see Table 6), it appears that the difference is hardly significant.

The need for a careful analysis of the value of sprays for the control of this disease indicated the necessity of making careful counts not only of the total number of blossom clusters affected but more specifically of percentages of primary or initial number of blossom clusters blighted per tree. Since secondary blossom blight originates very largely as internal bacterial extensions from the initial infections and, therefore, not controllable by external spray applications, there is the possibility of introducing considerable error in evaluating the controlling influence of sprays if reliance is placed entirely on counts of total numbers blighted. It is obvious that, when comparing trees having large numbers of blossom clusters on individual shoots with other trees in the same orchard having smaller numbers of blossom clusters per shoot, there may be a smaller number of initial infections and yet total counts might readily reveal greater numbers of blossoms blighted in case there are more blossoms per shoot, irrespective of the treatment given.

For this reason an attempt was made to make as accurate counts as possible of percentages of initial infections per tree. As the number of blossom clusters per tree was large and there was a relatively large number of trees, it was physically impossible to count all the clusters. Therefore, on any tree having a great number of blossoms, 400 clusters chosen at random over the whole tree were counted, and of these, the number found blighted was recorded. There were doubtless some errors made in sampling and it is almost certain that in some instances secondary infections were included among the initial ones, and

TABLE 6. PERCENTAGES OF INITIAL BLOSSOM BLIGHT, LEAZER'S ORCHARD, NEAR ROGERS, ARKANSAS. MAY, 1935

Tree number	Row 5	Row 4	Row 3	Row 2	Row 1
	Lime sulphur including two open-blossom sprays	Bordeaux including two open-blossom sprays	Bordeaux without two open-blossom sprays	Copper phosphate without two open-blossom sprays	Copper phosphate including two open-blossom sprays
	Per cent	*Per cent*	*Per cent*	*Per cent*	*Per cent*
1	12.25	2.75	7.29	6.75	10.25
2	13.00	—	7.17	20.25	21.75
3	11.75	6.50	5.75	14.00	19.25
4	6.00	5.21	8.54	12.25	—
5	19.50	5.00	6.59	9.50	7.75
6	8.02	—	6.50	11.00	9.75
7	23.25	6.50	8.00	—	—
8	18.00	—	15.25	15.50	10.62
9	24.25	3.10	—	14.25	1.90
10	10.59	0	14.25	16.00	14.34
11	3.95	8.25	4.35	5.59	8.66
12	7.55	7.25	10.00	7.62	0.96
13	11.75	4.73	7.00	10.24	9.25
14	9.75	7.25	11.75	4.46	—
15	9.06	4.32	4.42	10.75	8.38
16	8.49	9.00	6.06	8.49	28.57
17	no data	8.33	22.28	10.39	13.18
18	23.08	11.11	17.80	5.17	0.95
19	19.50	11.00	—	6.67	0.67
20	14.28	10.29	3.22	—	—
21	9.64	7.03	8.50	6.70	4.00
22	8.39	7.25	15.73	9.00	5.25
23	23.25	11.50	22.22	6.00	9.75
24	20.97	9.25	13.96	9.09	—
25	14.00	10.11	22.10	8.47	20.35
26	—	11.75	15.79	10.50	0
27	—	4.81	0	16.00	1.58
28	—	—	—	7.75	5.00
29	—	—	16.67	6.56	16.00
30	—	8.25	11.86	8.75	—
31	—	10.25	21.00	0	—
	ΣX 330.27	190.84	314.05	277.70	238.16
	MX 13.761250	7.340000	11.631481	9.917857	9.923333
	ΣX^2 5418.8421	1625.5996	4587.2775	3139.2918	3596.0592
	$(\Sigma X)MX$ 4544.928367	1400.765600	3652.86608	2754.18916	2363.340987
	$\Sigma X - (\Sigma X)MX$ 873.914063	224.8340	934.161142	401.102633	1232.718213
	$\sqrt{\Sigma X^2-(\Sigma X)MX}$ 29.562038	14.994466	30.564050	20.027554	35.110087
$M=\frac{\sqrt{\Sigma X^2-(\Sigma X)MX}}{n-1}$	1.231752	0.576710	1.132002	0.715270	1.462920

TABLE 6. PERCENTAGES OF INITIAL BLOSSOM BLIGHT, LEAZER'S ORCHARD, NEAR ROGERS, ARKANSAS. MAY, 1935—(*Continued*)

Comparison between rows	M. D.	σM. D.	$\frac{\text{M. D.}}{\sigma\text{M. D.}}$
5 - 4	6.2725	1.3661	4.59
5 - 3	2.2620	1.6924	1.34
5 - 2	3.8434	1.4466	2.66
5 - 1	4.2344	1.8106	2.34
4 - 3	4.0104	1.2507	3.21
4 - 2	2.4291	0.8901	2.73
4 - 1	2.0381	1.4064	1.45
3 - 2	1.5813	1.3381	1.18
3 - 1	1.9724	1.7252	1.14
2 - 1	0.3910	1.4847	0.26

Levels of Significance

Level	Odds	$\frac{\text{M. D.}}{\sigma\text{M. D.}}$
Barely significant	19 :1	1.96
Highly significant	99 :1	2.58
Infinitely significant	1½ Billion :1	5.40

initial infections as secondary, although a concerted effort was made to keep these unavoidable errors as low as possible by making several counts during the period in which initial infections appeared, and by having different individuals make independent counts of the same row.

Table 6 shows quite clearly that, when comparing the lime-sulphur check row with the one sprayed with Bordeaux mixture which included the two open-blossom sprays, the odds are far greater than 1,000 to 1 that the difference between the percentages of blight in these two treatments is significant. Similarly, there is a very high level of significance in the different percentages of initial blossom blight shown by those Bordeaux-sprayed trees that received two open-blossom sprays when compared with those that lacked the two open-blossom sprays. Indeed, while the percentage of disease is lower in the row sprayed with Bordeaux mixture lacking the two open-blossom sprays than in the check row sprayed with lime sulphur, statistical analysis reveals no significant difference between the two. In other words, the absence of two open-blossom Bordeaux sprays in

the Bordeaux-sprayed row results in no better control of blossom blight than the lime sulphur sprays. Of course, it would have been desirable to have had a row without any spray but this being a privately-owned commercial orchard, this was found to be impractical. However, since many years of observation on fire blight in orchards which regularly receive lime sulphur sprays have shown no control by these sprays, the need for an additional check row having no sprays is not so evident.

Comparing the copper phosphate sprays with the Bordeaux sprays, the data admit of no conclusion. While the mean percentage of blossom blight is greater on the phosphate-sprayed trees, including those with two open-blossom sprays, than the comparable Bordeaux-sprayed trees, statistical analysis revealed no significant difference. The standard deviation of the mean being greater in the copper phosphate row, which included two open-blossom sprays, than in any other row, comparisons between this row and any other one become especially difficult to interpret properly.

It is concluded that the two Bordeaux sprays, applied first when 20 to 25 per cent of the blossoms were open, and second when 75 per cent of the blossoms were open, reduced the percentage of blossom blight very materially. The other Bordeaux sprays seem to have had very little or no influence on the amount of blossom blight, and, so far as the 1935 season is concerned, may well have been omitted or replaced by lime sulphur. This conclusion is especially warranted for that year because twig blight, which might be controllable by other Bordeaux sprays, was insignificant in amount.

The fact that the Bordeaux blossom sprays yielded only about a 50 per cent control of the disease was not at all surprising considering the fact that the blossoming period in 1935 was so irregular on the different trees that no two trees had anything near a comparable number of blossoms open at any one time. From a practical point of view, the difference between the cost of the sprays and the amount of control effected was such that the sprays appeared to be highly economical. No exact figures on the actual saving can be given because of the great variation in number of blossom clusters borne in this season on the different trees, and the variation in size of the trees.

As to the influence of the different sprays on the setting of fruit, here also the data were too irregular to permit exact evaluation. While careful counts were made of the number of drops occurring in the different treatments in May and June, the figures were of questionable value because of the great disparity in the original number of blossom clusters on the trees. It may be recorded, however, that there was no indication of reduction in

fruit setting in either the Bordeaux-sprayed rows or in the copper phosphate rows when compared with the lime-sulphur sprays. The figures suggested a proportionately greater amount of dropping of fruit on the lime-sulphur row.

Counts of the amount of spray injury, including russeting and burning of fruit, were made in the various treatments, as shown in Table 7. It is well known that the amount and severity of spray injury induced by various kinds of sprays on apples is intimately related to weather conditions. During cool, wet weather in the early part of the growing season, various sprays are known to induce russeting of fruit, Bordeaux mixture in particular being notorious in this respect. Table 7, taken in conjunction with the records of temperature and rainfall shown in Figure 9, reveals this quite clearly.

Out of the 8 years in which the writer has been applying weak Bordeaux sprays to various apple orchards in the Ozarks of Arkansas in the early part of the growing season, the 1935 season was by far the most conducive to the development of russeting. Even lime-sulphur sprays, which ordinarily produce little or no such injury on apples in the early season, caused an appreciable amount of russeting in this year. If careful grading of the fruit were practiced, 66.9 per cent of the fruit shown in Table 7 to be severely russeted in the Bordeaux-sprayed row without two open-blossom sprays and 84.5 per cent of the Bordeaux-sprayed row with two open-blossom sprays, would have been discarded from any first-grade pack. Indeed, even a considerable number of those included among the apples showing slight spray injury would not have gone into any fancy pack.

TABLE 7. SPRAY INJURY ON APPLES AT LEAZER'S ORCHARD, NEAR ROGERS, ARKANSAS. FALL OF 1935

Row number	Type of spray	Apples examined	Apples showing slight spray injury	Apples showing excessive spray injury	Apples with excessive spray injury
		Number	*Number*	*Number*	*Per cent*
1	Copper phosphate with two open-blossom sprays	1,810	665	573	31.6
2	Copper phosphate minus two open-blossom sprays	654	259	254	38.8
3	Bordeaux mixture minus two open-blossom sprays	1,268	391	849	66.9
4	Bordeaux mixture with two open-blossom sprays	1,580	232	1,336	84.5
5	Lime sulphur with two open-blossom sprays	723	243	95	13.1

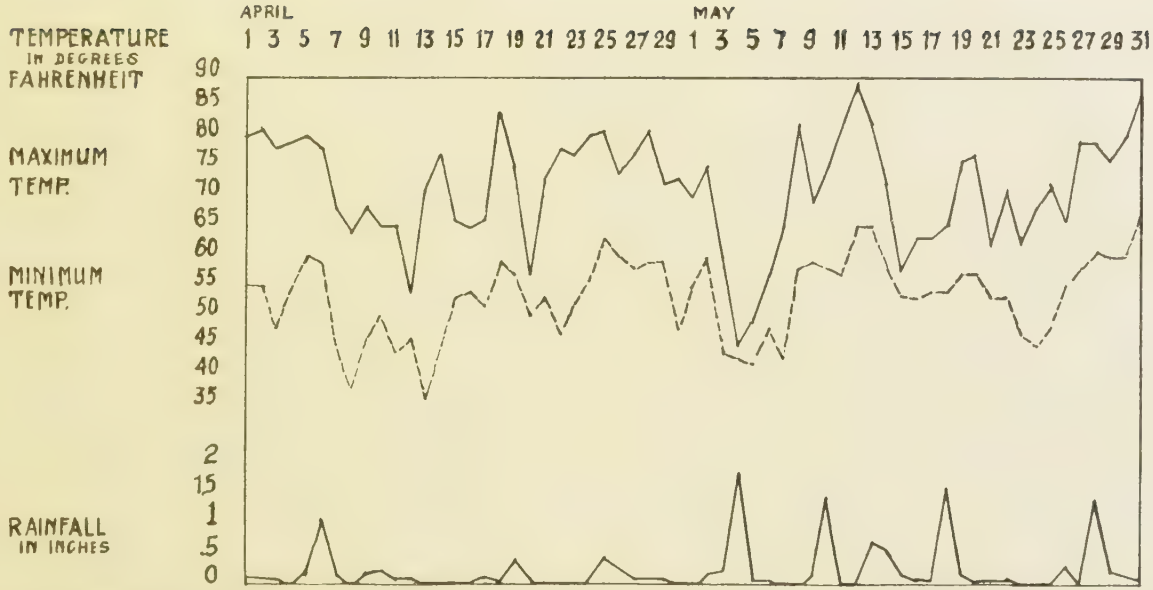

Figure 9. Climatological data for the region near Rogers, Arkansas, during the period of blooming and of maximum fire-blight development in the orchard used for spray date investigations.

While there is good evidence to indicate that a considerable reduction in amount and severity of spray injury might be expected if the sprays designed to control blossom blight excluded the cluster bud and calyx sprays, there can be no doubt that the addition of two open-blossom Bordeaux sprays will result in some russeting, given cool, wet weather. A comparison between Rows 3 and 4 indicates that these two sprays were responsible for an increase of 17.6 per cent in number of apples showing severe spray injury in 1935. Table 7 also shows that there was more than twice as much fruit with severe russeting on the Bordeaux-sprayed rows than on those sprayed with copper phosphate. If the latter spray were as economical as Bordeaux, and as effective in controlling blossom blight and apple scab, it might readily be substituted for Bordeaux mixture. Unfortunately, the records do not indicate this. Furthermore, aside from the question whether it controls fire blight as well as Bordeaux mixture, under a combination of weather conditions such as existed in 1935, it causes far more injury to the fruit than lime sulphur sprays.

A comparison of the percentages of severe russeting of copper phosphate-sprayed Rows 1 and 2, with and without two open-blossom sprays, shows a marked discrepancy in favor of the row which received the two additional sprays. The figures, however, are of questionable significance since the number of fruit examined in Row 2 was relatively small. Had it been possible to examine as many fruit on Row 2 as on Row 1, there is little doubt that the percentages would have been either equal or in reverse form from that shown in Table 7. So far as concerned the general appearance of the fruit as they still hung on the trees shortly before being picked in late August, there was no noticeable difference between Rows 1 and 2.

The weather records, shown in Figure 9, were taken from Climatological Data of the U. S. Department of Agriculture Weather Bureau, Arkansas Section. Since the Bentonville station, where these records were obtained, is approximately 3 miles away from the Leazer orchard utilized for spray investigations, they are quite applicable to this orchard.

It will be noted that the temperatures for the two months were exceptionally low, especially during the blooming and fruit-setting period of Jonathans which, taken from the late cluster bud stage to petal fall, lasted from April 9 to May 7. Not only were the minimum temperatures low for two months, the mean temperatures were equally low and below normal. While the precipitation was below normal in April, there were almost daily rains throughout the latter part of the blooming period. It is this

combination of low temperatures and excessive precipitation which is known to be conducive to spray injury.

In summarizing the study of russeting induced by sprays, it may be said that for those apple regions which regularly aim to market fancy packs, there is little doubt that in cool, wet seasons, the amount of russeting induced by one or two full-blossom Bordeaux sprays will be considerable and, therefore, seriously reduce the amount of No. 1 fruit. On the other hand, in the Ozarks of Arkansas and Missouri, and in any other region where the grading of fruit is not very strict and where much of the fruit is sold in bulk, the amount and degree of russeting is of little economic importance. Despite the severe russeting experienced in 1935 with Bordeaux sprays, as seen in Table 7, the grower received the same price for these fruits as for the smoother and better colored fruit of the lime-sulphur row.

While it would be highly desirable to find a substitute for Bordeaux mixture so as to reduce the amount of russeting, or to combine it with other materials which might lessen its toxicity to apple tissue, and experiments are underway with that end in view, nevertheless, so long as the price of apples remains as low as it is, there is no particular reason for withholding Bordeaux sprays to control blossom blight wherever the disease is serious in the Ozarks. Furthermore, aside from the control of blossom blight occasioned by such sprays, the problem of obtaining good setting of fruit in this region may partly be solved by the use of such sprays, as will be discussed later.

Comparison of Spray Investigations for Fire-Blight Control in 1933, 1934, and 1935

Including the spray investigations of 1932, presented in a previous publication *(26)*, data are now available for 4 successive years of spray investigations for the control of fire blight, all in fair-sized commercial orchards. For 1932 and 1933 these involved an isolated apple orchard near Ozark, Arkansas, while for 1934 and 1935 they included an orchard located in a predominantly apple-growing area near Rogers.

In the first orchard, fire blight had been far more abundant as a whole than in the second, and in each of the 2 years in which the investigations were conducted the control of the disease by timely Bordeaux sprays was very marked. In the second orchard, the disease has appeared sporadically and not generally so abundant. In the latter orchard, the control of the disease was not as thorough as in the first; in the season of 1934, when fire blight was of relatively minor importance in this orchard, it is

doubtful if the two additional open-blossom sprays were of sufficient value to justify their use for the control of blossom blight. However, so far as the setting of fruit was concerned, there was considerable indication that the full-blossom sprays were helpful. Even in 1935, when more disease appeared in this orchard than in the previous year, the control was not nearly as perfect as in the Ozark orchard, though, from an economic viewpoint, it was doubtless of value.

The conclusion is inescapable that spraying for the control of fire blight, particularly for blossom blight, is not as effective in some seasons as in others; however, spraying for the control of fire blight probably does not differ greatly from the variation in control of various other parasitic apple diseases by the use of sprays. The writer's work on the control of blossom blight of apples by the use of weak Bordeaux sprays in large commercial orchards has been at least partly confirmed by several other investigators, including Musser *(18)* in South Carolina, Niswonger *(19)* in North Carolina, and Andes *(1)* and Prather *(21)* in Tennessee. Likewise, McCown's *(14, 15, 16)* investigations in Indiana, largely dealing with the control of fire blight by the application of one full-bloom spray on artifically infected apple blossoms, though records are also given of a few naturally infected apple and pear trees, are in line with the writer's findings; however, the remarkable degree of control of blossom blight McCown has obtained by the use of one Bordeaux spray has not been obtained consistently by the writer even with several early season Bordeaux applications. As with any new remedy, there is danger of over-emphasizing the importance of sprays for the control of fire blight.

While the work up to the present indicates that Bordeaux sprays are not only of value for the control of blossom blight but possibly also for a proper setting of fruit, such sprays cannot be recommended as substitutes for lime sulphur in varieties that are particularly susceptible to scab; and in any region where fire blight and pear and apple blast are not serious, and where russeting is a serious economic factor in the grading of fruit, it is even a question if the Bordeaux blossom sprays might not do more harm than good. However, for the main apple growing region in Arkansas and for the Ozarks of Missouri, the evidence appears sufficient to indicate that two open-blossom applications of 1-3-50 Bordeaux mixture applied, first, when approximately 25 per cent of the blossoms are open, and, second, when 75 to 80 per cent are open, are of unquestionable value in the control of blossom blight. Depending upon the timeliness of the spray applications to the dissemination of the bacteria and in relation to weather

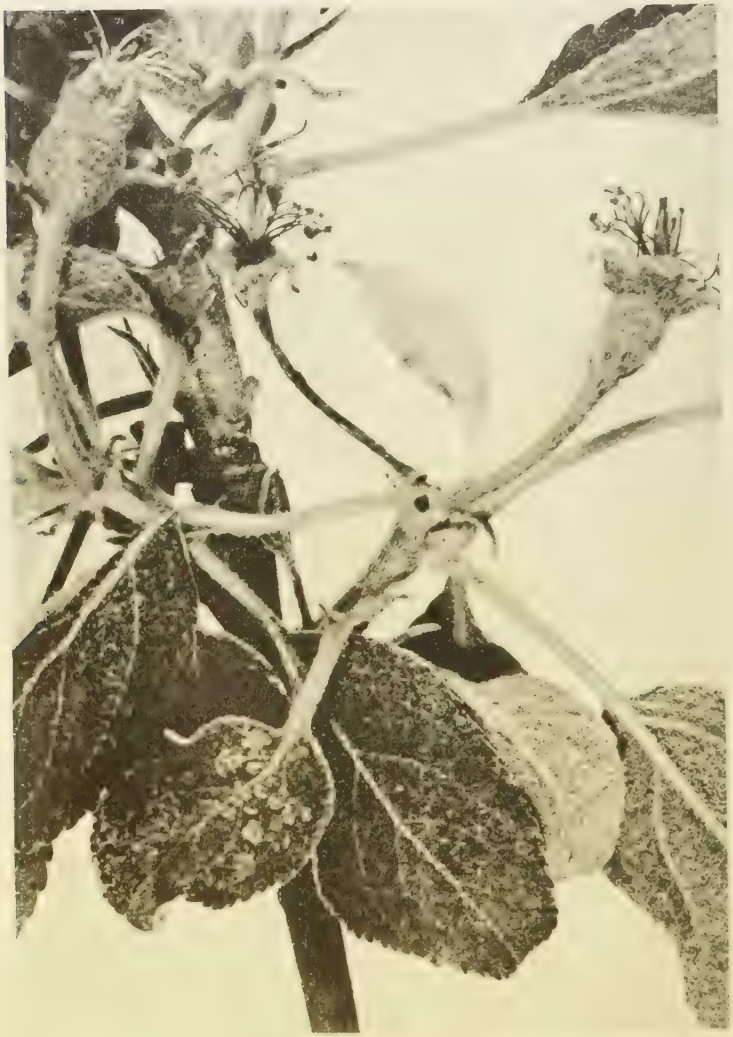

Figure 10. A Jonathan blossom, toward the center, affected by fire blight and abscissed from the original cluster. The material was gathered May 9, 1934, in the orchard used for spray investigations. Such abscission has been found to be of common occurrence. Photographed May 9, 1934

conditions, the control of blossom blight, and to a large extent secondary twig blight, will be more or less thoroughly obtained.

Though winter pruning of all dead and diseased wood is highly desirable from various points of view, such pruning, no matter how carefully conducted, has not been found to serve as a substitute for weak Bordeaux sprays in the control of the blossom blight stage of fire blight. While canker treatments, including Day's zinc chloride applications, have not been tried by the writer, economically they are of questionable value for the control of this disease on apple trees in Arkansas. Up to the present no evidence has been found which would indicate that such cankers serve as an important source of inoculum for primary blight in Arkansas.

ABSCISSION OF PEAR AND APPLE BLOSSOMS IN RELATION TO INFECTION BY ERWINIA AMYLOVORA AND PHYTOMONAS SYRINGAE

One of the symptoms commonly associated with fire blight concerns the tenacity with which attacked leaves, blossoms, and fruits remain attached to the trees. Occasionally, however, there has been noted, during the past few years *(26)*, aborted and abscissed apple blossoms, which were affected with fire blight, clinging loosely to the cluster during the flowering period (Figures 10 and 11). Often the external symptoms were obscure and could only be recognized by the occasional presence of droplets of sticky bacterial exudate. During the blossoming period of Kieffer and Garber pears in 1935, many blossoms that were still clinging to the spurs were found showing various sized blackish spots on the receptacle cups, and also at times showing a blackish discoloration of styles (Figures 12 and 13). These spots often peppered the whole of the cup, although many blossoms showed only one or two small localized discolored areas. In addition to such discolorations, there was also noted in numerous instances a marked, water-soaked appearance of portions of the

Figure 11. Two Jonathan blossoms which had abscissed and were found hanging loosely on a twig; the one to the left was infected with Er. amylovora this pathogen. Gathered on May 10, 1934, in the

Figure 12. Kieffer pear blossoms showing dead blackish spots in the receptacle cups from which **Er. amylovora** and **Phytomonas syringae** were isolated. Such blossoms invariably abscissed. Gathered on April 6, 1935, in one of the orchards used for studying the oversummering of **Er. amylovora** in 1934. Some trees which had large numbers of such infected and abscissed blossoms later on showed no other fire blight or blast symptoms.

fleshy receptacle, suggesting a common symptom of fire blight. However, in many blossoms which showed blackish spots in the cup there was no accompanying water-soaked appearance of the adjoining receptacle tissue. These trees had received no fungicidal or bactericidal sprays.

A few days after the spots were detected, such blossoms began to abort and drop from the trees. In the two pear orchards in which close studies were made in 1935, the number of blossoms

Figure 13. Abscissed Kieffer pear blossoms affected with blast and fire blight. They were gathered on April 18, 1935, near Bentonville, Arkansas. Fully 85 per cent of the abscissed blossoms showed infection with **Er. amylovora** or **P. syringae**, or both.

Figure 11. Typical fire-blight symptoms induced by inoculating tender Bartlett pear shoots with single-colony transfers of cultures isolated from abscissed pear blossoms having blackish receptacle cups. Photographed April 29, 1935, four days after the shoots were inoculated.

showing such symptoms was so great and the abscission so common that the number of blossoms remaining was insufficient to produce a crop adequate to the age and size of the trees. Not all of the abscissed blossoms showed such symptoms, though the proportion was high, and amounted to approximately 85 per cent of the blossoms that had dropped. Since the weather during the

Figure 15. Typical pear blast symptoms induced by inoculating tender Bartlett pear shoots with single-colony transfers of cultures isolated from abscissed pear blossoms having blackish receptacle cups. Photographed April 29, 1935, four days after the shoots were inoculated. (Compare with Figure 14.)

blossoming period was characterized by excessively cool and partly rainy days, it has been assumed that the small amount of fruit set in the 1935 season in the Ozarks was due to improper pollination, and while this factor doubtless was operative in some of the blossoms, it seems evident that other factors were of equal or greater importance. What were these factors?

As reported in a previous publication *(28)*, a high percentage of the blossoms which possessed blackened receptacle tissues showed bacteria present within the tissues. Numerous isolations were attempted and pathogenic bacteria were obtained in pure cultures in over 70 per cent of the trials. As revealed in artificial infections on succulent Bartlett pear shoots, two different species of bacteria were involved, one the fire-blight pathogen, *Erwinia amylovora*, and the other the pear-blast organism, *Phytomonas syringae*. Out of 64 isolations from abscissed Kieffer and Garber pear blossoms gathered in two counties, 46 yielded pathogenic bacteria, 29 of which were typical *Er. amylovora* and 17 *P. syringae*, as revealed in artificial inoculation tests. In some of the affected blossoms, both types of bacteria were present.

It was found to be exceedingly difficult to distinguish between the two species when colonies of both were present in any one series of poured dilution plates, using dextrose beef-extract agar as the medium. One of the simplest means of identifying them was by utilizing hypodermic injections, into tender Bartlett pear shoots, of bacterial suspensions in water made from single-colony transfers. Using this simple inoculating technique, the fire-blight bacteria isolated from the abscissed blossoms produced symptoms characteristic of fire blight (Figure 14), while the pear-blast bacteria similarly inoculated produced somewhat different symptoms (Figure 15). Note the more localized stem infections and particularly the discoloration of the leaves. This intense discoloration is sometimes greater between the veins, and at other times greater along the veins. Unlike the fire-blight bacteria which travel into the leaf tissues and can readily be found in any part of the discolored areas in fresh infections, the pear-blast bacteria have not been found in the discolored leaves attached to the inoculated stems.

One of the most interesting things observed on trees showing large numbers of abscissed blossoms affected with fire blight, with blast, or with both, was the more or less complete absence of fire blight or blast symptoms on twigs, limbs, trunks, or leaves throughout the growing season. Indeed, aside from the young blossoms, the trees as a whole were so free from fire blight and blast that, if the blossoms had not been carefully studied, there would have been no reason to suspect that these diseases had

been at all prevalent in that season. This suggests, among other things, the need for a closer bacteriological and, possibly, also mycological study of abscission in pear and apple blossoms in localities where a proper setting of fruit is a problem, and that such studies need not be confined to varieties or even species that are presumably susceptible to fire blight or to blast. In any case, it seems clear that the great reduction in fruit set, associated with these two diseases in 1935, calls attention to a phase of fire blight and blast injury that has largely been overlooked in the past in this country.

In the British Isles, Barker and Grove *(2)* first called attention to the abscission occasioned by attacks of a whitish fluorescent species of bacteria which was later named *Bacillus Barkeri (3)* and which, without much doubt, belongs to the group typified by *Phytomonas syringae (27)*. In South Africa, Doidge *(6)* in 1917 and DuPlessis *(7)* recently reported a dropping of pear blossoms caused by attacks of *Bact. (P.) nectarophilum*, another whitish fluorescent organism closely related to *P. syringae (27)*.

In the accounts of the various spray investigations presented previously for the control of fire blight, it has been noted that apple trees sprayed with a weak Bordeaux mixture when the blossoms are wide open often showed a superior setting of fruit to those not receiving such sprays, though no adequate counts were made. This has always been somewhat surprising because one would expect such a caustic material to interfere to some extent with pollination; in fact MacDaniels, Hildebrand, and Burrell *(12)* have found that very weak Bordeaux mixtures seriously reduce pollen germination and also, to some extent, setting of fruit. However, in view of the finding that *Er. amylovora* and *P. syringae* are to be found in some seasons in a very high percentage of abscissed blossoms, it would seem that open blossom sprays possessing germicidal effects on these two organisms may be highly beneficial even though they may hinder pollination to some extent. In other words, comparing any possible retarding or caustic effect on either stigma or pollen grain by Bordeaux sprays with the effects on the blossoms caused by infection with these pathogens, the latter seem to be far more important in causing abscission or in preventing the setting of fruit than the former.

It will be noted in another publication that so far as the fire-blight organism is concerned, it has been experimentally demonstrated that the stigmatic surface forms one of the most receptive avenues for infection. This probably is also true for the blast organism in view of the fact that it has been isolated from

naturally infected, discolored styles. Once these bacteria gain entrance into the stigma, their agressiveness and rapidity of invasion are such that pollen tubes can hardly be expected to compete with them in reaching the receptacle. There is even a suggestion that other types of microorganisms, including various fungus spores, upon lodging on the stigma can, at least at times, render the stigmatic surface unreceptive to pollen-tube penetration.

The writer has often noted fungus growth on both pear and apple stigmas of relatively young blossoms. However, the influence of such growth on the production of an abscission layer remains to be determined. Likewise, there is as yet no experimental evidence to explain why *Er. amylovora* will, in some instances, invade not only a whole blossom but pass into the spur and even the main branch, while in other instances the invasion remains much more localized and confined to the upper parts of the receptacle. *P. syringae*, on the other hand, nearly always confines its invasion to a limited amount of tissue, whether it be twig, leaf, blossom, or fruit, and from this viewpoint is somewhat more insidious than the fire-blight pathogen since it is more apt to be overlooked.

It seems proper to conclude from these investigations of abscissed pear and apple blossoms infected with *Er. amylovora* and with *P. syringae*, that weak Bordeaux sprays applied to open blossoms are not only of value in controlling blossom blight, but what may eventually prove of greater importance, such sprays may materially help to obtain a good setting of fruit.

A STUDY OF THE GERMICIDAL ACTION OF VARIOUS COPPER SPRAYS ON ERWINIA AMYLOVORA

In 1929, McCown (*14*) determined, by placing *Er. amylovora* in liquid suspensions of Bordeaux mixture in test tubes and on Bordeaux films dried on glass slides, that very dilute mixtures of this spray material possess marked germicidal action on these bacteria. It seemed desirable to duplicate his work, to use more refined quantitative methods, and also to determine the possible germicidal effects of various other copper sprays. For this purpose the following experiments were conducted:

Experiment No. 1. The influence of copper sulphate on the growth of the fire-blight pathogen. This experiment was primarily designed to determine if the copper salt used in Bordeaux mixture has any detrimental action on the growth of this organism. To determine this, a sufficient amount of copper sulphate was added to dehydrated potato dextrose agar so as to give a

concentration equivalent to that in 1-3-50 Bordeaux mixture. Pure culture transfers were then made to tubed slants of this medium and also to the same medium lacking the copper salt. No growth was obtained in any of the slants containing copper sulphate, while those free from the copper all yielded excellent growth. The hydrogen-ion concentration of both kinds of media was pH 5.6. It was found that while the copper salt completely inhibited the growth of *Er. amylovora*, it did not prevent a good growth of another plant parasite, *Sclerotium rolfsii*. Evidently, copper sulphate is very toxic to the fire-blight organism in a vegetable medium which is otherwise highly conducive to its growth.

Experiment No. 2. The influence of Bordeaux mixture, copper oxide, and copper phosphate spray mixtures on the viability of the fire-blight pathogen. To numbered and weighed glass slides, a spray of Bordeaux mixture, copper oxide mixture, and copper phosphate mixture was applied, the slides being held at the same distance, and sprayed approximately the same length of time. The Bordeaux mixture utilized was the weak 1-3-50 formula, while the copper oxide and copper phosphate followed Goldsworthy and Green's (*8*) formula of 2 pounds of the respective copper salts, 4 pounds hydrated lime, 2 pounds bentonite to 50 gallons of water.

After being sprayed, each slide was placed in a sterile petri dish and allowed to dry at room temperature. Having come to a constant weight, all slides of the three different sprays, which possessed a comparable quantity of dried spray material, were then used for germicidal tests. The amount of dried spray material on the slides used was 5 mgs. From a 3-day-old nutrient agar slant of *Er. amylovora* a small amount of bacterial mass was transferred to a tube of 10 cc. of sterile water; likewise, from a 5-day-old nutrient broth transfer of the same organism, but representing another isolate, a small drop of bacterial suspension was added.

From this water suspension a 3 mm. loopful of bacteria was placed in three separate places on each dried film of spray and permitted to remain for 10 minutes and for 30 minutes. At the end of each period a 2 mm. loopful was transferred to 0.1 cc. sterile water contained in a petri dish and melted nutrient dextrose agar added and stirred with the water. Suitable checks were maintained by transferring comparable quantities of bacterial suspensions to sterilized glass slides having no sprays.

At the end of 3 days when all the colonies were fully visible, they were counted as shown in Table 8.

TABLE 8. AVERAGE NUMBER OF COLONIES OF *Er. amylovora* RESULTING FROM APPLICATIONS OF DIFFERENT SPRAY MATERIALS

Type of spray film	10 minutes	30 minutes
	Number	*Number*
Bordeaux mixture 1-3-50	0	0
Copper oxide-lime-bentonite 2-4-2-50	1,986	1
Copper phosphate-lime-bentonite 2-4-2-50	0	0
Check	9,898	5,731

This experiment indicates that, given equal weights of dried Bordeaux film and copper phosphate film, 5 mgs. each, these sprays are completely lethal to a population of 9,898 bacteria within 10 minutes, while a similar weight of copper oxide film still leaves a high percentage of bacteria alive. Even at the end of 30 minutes the copper oxide mixture is not completely lethal to the bacterial population utilized, the one colony shown in Table 8 proving, on subsequent transfers into nutrient broth and inoculation into tender Bartlett pear shoots, to be just as virulent as check transfers.

Experiment No. 2 was duplicated and the results obtained were essentially the same as those presented here.

Experiment No. 3. An effort to determine the toxicity of copper phosphate mixture to a large population of Er. amylovora. As there was a suggestion in the experiments cited previously of a more or less direct relationship existing between the number of bacteria introduced on a given amount of dried spray and the number of bacteria surviving, it seemed desirable to test the influence of a known quantity of copper phosphate mixture on very large numbers of bacteria. In order to determine the possibility of such relationship, copper phosphate mixture was sprayed on glass slides and the technique used previously was followed with the following modifications: a measured volume of bacterial suspension was used and the checks were diluted with known quantities of sterile water so as to permit a more accurate counting of colonies; three series of dried spray films of different weights were used in order to determine the toxicity of varying quantities of spray to a given number of bacteria; and different quantities of bacterial suspension were applied to each slide so as to permit sampling of equal quantities at different time intervals. Serological pipettes of high accuracy were used for the

last purpose. As in the previous tests, triplicates were made of each series and of each time interval, and the bacterial counts represented the average of three plates. The results are shown in Table 9.

Except for comparative value, the figures in Table 9 are of no particular significance. Although a concerted effort was made to avoid errors in sampling and in manipulation, the variation in any one series was so great with reference to number of colonies that no high degree of accuracy can be claimed. The main point for which there is ample evidence is that, given a certain quantity of dried spray material under comparable laboratory conditions of light, temperature, and moisture, the complete killing of all bacteria applied to the dry film depends on at least two factors, namely: the number of bacteria applied, and the length of treatment. In other words, in the presence of a droplet of water containing a very large number of bacteria, such as may be found in a partly dissolved droplet of exudate under field conditions, a spray film may not be lethal to all the bacteria in a given time interval, and if this spray film is partly removed or washed away by rain, its protective action is doubtless lessened. This, in turn, signifies that even in the presence of a protective coat of spray the control will not be effective in all instances. The results here reported on the inability of a given quantity of bactericide to kill fire-blight bacteria when the number is exceptionally large are very comparable to those obtained by McCallan *(13)* and Rippel *(23)* who found that with various fungi, the toxicity of a given quantity of disinfectant depends, in part, on the spore numbers.

TABLE 9. AVERAGE NUMBER OF COLONIES OF *Er. amylovora* RESULTING FROM TREATMENTS WITH COPPER PHOSPHATE MIXTURE

Length of treatment	Dry weight of spray film			
	2.3 mgs.	2.5 mgs.	3.0 mgs.	Check
	Number	*Number*	*Number*	*Number*
5 minutes	10,617	9,303	7,726	
10 minutes	8,791	16,598*	6,523	
20 minutes	7,939	6,590	2,757	
30 minutes	1,827	564	117	12,330,000

*Probably an error in manipulation.

Experiment No. 4. Determining the toxicity of two different copper sprays with and without arsenate of lead. As arsenate of lead has been applied in combination with Bordeaux

mixture and copper phosphate mixture in the calyx spray and first cover spray in field experiments for the control of fire blight, it seemed desirable to determine if the arsenical had any influence on the germicidal action of copper sprays against *Er. amylovora*. Likewise, it appeared worthwhile to recheck the germicidal action of copper oxide mixture which, as shown in Experiment No. 1, was rather weak in its action. The amount of arsenate of lead added was in the proportion of 1½ pounds to 50 gallons of spray.

The same technique described previously was used, and the number of bacteria developed at the end of 3 days for the various treatments was counted as shown in Table 10.

Owing to the variation in the weights of the dry spray films in the different treatments and in the different time intervals of any one treatment, the counts naturally vary and are not very consistent with the toxicity expected in the various time intervals of treatment. Had it been possible to use only slides which had the same amounts of dry spray film, the counts of colonies would doubtless have been more consistent with the length of treatment. Nevertheless, the figures unmistakably reveal a lack of lethal action of the copper oxide mixture, particularly the combination with arsenate of lead. There is actually a marked reduction in toxicity of this combination when compared

TABLE 10. AVERAGE NUMBER OF COLONIES OF *Er. amylovora* RESULTING FROM TREATMENT WITH COPPER PHOSPHATE AND COPPER OXIDE MIXTURES WITH AND WITHOUT ARSENATE OF LEAD

Type of dry spray film	Weight of dry spray film	Length of treatment			
		5 min.	10 min.	20 min.	30 min.
	Mgs.				
Copper oxide mixture	2	9			
	1.1		165		
	1.9			28	
	1.3				29
Copper oxide mixture plus arsenate of lead	1.4	1,084			
	1.1		966		
	1.2			729	
	1.2				325
Copper phosphate mixture	5.0	44			
	5.0				
	6.0			0	
	7.0				
					0
Copper phosphate mixture plus arsenate of lead	9.0	0			
	9.0		0		
Check					3,943

with the same copper salt mixture without arsenate of lead. However, it is very doubtful if this is to be attributed to the arsenical. The probable explanation for this is as follows: The copper oxide used possessed a marked tendency to precipitate out of suspension rather rapidly, and, when it was combined with the lead arsenate, (the latter salt remaining in suspension better than the copper oxide), replaced the copper salt to some degree. Thus, a dried film containing the oxide and arsenical would have less copper per given weight of dried spray than the same spray without the arsenical.

With reference to the copper phosphate mixture, with and without arsenate of lead, the most noteworthy result was the survival of three colonies in each of the 20 and 30-minute treatments where the arsenical was present. This, however, is probably correlated with the fact that the arsenical partly replaced some of the copper salt and not because of any inherent reduction of the germicidal action of the spray due to the presence of arsenate of lead.

The conclusion seems warranted that arsenate of lead does not increase or decrease the germicidal action of copper oxide and copper phosphate mixtures to *Er. amylovora*.

SUMMARY AND CONCLUSIONS

Studies of oversummering of fire-blight bacteria within pear twigs and limbs infected in the early part of the growing season show a gradual reduction in the number of cases in which the bacteria are still viable as the summer advances. In 1934 the number of infected twigs and limbs with viable and infectious bacteria in July constituted somewhat less than 2 per cent of the cases studied. This was reduced to 0.78 per cent in August. In September, out of 165 blighted twigs and limbs studied, none yielded viable cultures of *Erwinia amylovora*.

It is concluded that blighted twigs and limbs of both pear and apple varieties commonly grown in Arkansas are not nearly so important in serving as sources of inoculum for the succeeding years fire blight as in regions of shorter growing seasons where other varieties are used, or as in certain pear-growing areas of Pacific states.

In attempting to determine any possible correlation between the amount of fire blight present on individual trees in one season of a susceptible apple variety with the amount present in the succeeding year, counts were made of all blighted branches, including blossom clusters and twigs and limbs present on each tree in the late winter and early spring and compared with

counts made on the same trees after new blight of the next season had fully developed. The counts involved 167 contiguous trees and were of large quantities of blighted material which had been permitted to remain on the trees through the winter and up to the next growing season. No correlation was noted between the amount of fire blight on individual trees in one season with the amount of the succeeding season.

The results of spray investigations for the control of fire blight, particularly blossom blight, in 4 successive years were compared and the details presented for 3 of these years. In 1933 control by means of 1-3-50 Bordeaux mixture applied (1) as a cluster bud spray, (2) when 25 per cent of the blossoms were open, (3) when 75 to 80 per cent of the blossoms were open, (4) as a calyx spray, and (5) as a first cover spray, was almost perfect, and the results were very similar to those obtained in the same orchard in 1932. In 1934, using another orchard but of the same variety (Jonathan), the results were inconclusive because of the relative scarcity of the disease throughout the orchard. In that year copper phosphate mixture, a new fungicide, was included in the control block as well as the Bordeaux mixture. In 1935, using the same orchard as in the previous year, a 50 per cent control of blossom blight was obtained by means of 1-3-50 Bordeaux sprays which included two open-blossom sprays over check trees sprayed with the usual lime sulphur sprays, and which included two open-blossom sprays of lime sulphur. Trees sprayed with Bordeaux mixture in the cluster bud stage, in the calyx stage, and in the first cover spray, but not in the open-blossom stages, showed no better control than the lime sulphur-sprayed check trees. Trees sprayed with copper phosphate mixture with and without two open-blossom sprays showed such irregularity in controlling the disease that no conclusions are warranted.

Evidence is presented indicating that in the 1935 spray investigations the two open-blossom Bordeaux sprays were responsible for the control of blossom blight and, indirectly, twig blight that was obtained.

Records kept of the amount of fruit russeting occasioned by early season Bordeaux sprays vary considerably from year to year. In most of the years for which data were obtained, the amount of russeting was negligible, while in 1935 it was exceptionally severe. However, this severity was of no economic consequence in the region involved in these investigations. For other apple-growing areas, particularly where fancy packs are used or very strict grading is followed, russeting to the extent obtained in the Ozarks of Arkansas in 1935 would seriously reduce the value of the fruit. For these other areas it is suggested that wherever

fire blight is a serious problem, 1-3-50 Bordeaux mixture, applied at least twice during the period when the blossoms are wide open, should be given a thorough trial and the amount of control carefully compared with any possible reduction in value of fruit due to spray injury.

Relative to the influence of early season Bordeaux sprays on the setting of fruit, no accurate data were obtained, although the records of the number of drops of Bordeaux-sprayed trees compared with those receiving lime sulphur sprays suggest that fruit setting is superior on trees sprayed with the weak Bordeaux mixture.

A study of abscission of pear and apple blossoms revealed that the fire-blight pathogen, *Er. amylovora*, and the pear-blast pathogen, *P. syringae*, were at times found in a high percentage of blossoms that had failed to set. In 1935, approximately 85 per cent of abscissed pear blossoms in two different counties showed symptoms indicating infections by one or both of these organisms, and the number of blossoms remaining on the trees was entirely inadequate to produce a fair-sized crop. Such infected and abscissed blossoms were found on trees which showed little or no other fire blight or blast symptoms throughout the remainder of the growing season. For this reason the importance of these pathogens in preventing a proper setting of fruit is likely to be overlooked. It seems possible that the better setting of fruit noted in the spray investigations aimed primarily to control blossom blight, may have been due in part to the control of abscission induced by *Er. amylovora* and *P. syringae*. These investigations suggest that even in orchards where there are no recognizable symptoms of fire blight or blast, two open-blossom Bordeaux sprays are of value in obtaining a better setting of fruit. Aside from their germicidal properties, dilute copper sprays possess something of a stimulatory action, while lime sulphur has a tendency to have the opposite effect, according to Dr. J. W. Roberts.[6]

Investigations concerning the germicidal action of various copper compounds on the fire-blight pathogen reveal the following: Copper sulphate, when used at the same strength as that in 1-3-50 Bordeaux mixture and incorporated in a vegetable medium which otherwise promotes excellent growth of these bacteria, completely inhibits all growth. Five mgs. of dried 1-3-50 Bordeaux film is completely lethal to a population of approximately 10,000 bacteria within 10 minutes. The same weight of dry film of copper phosphate mixture is also lethal to a similar number of bacteria, while a dry film of copper oxide mixture of the same weight is only toxic to about 80 per cent of such a popu-

[6]Personal communication.

lation within 10 minutes and is not completely lethal even in 30 minutes. When the bacterial population is greatly increased and the quantity of a dried film of copper phosphate mixture is slightly reduced, the toxicity diminishes within given time intervals. A comparison between copper phosphate and copper oxide mixtures with and without arsenate of lead indicates that the arsenical in combination with these sprays possesses no toxicity to *Er. amylovora*.

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